SCIENTIFIC NOTE

Survival of male click beetles, *Agriotes obscurus* L., (Coleoptera: Elateridae) during and after storage at different temperatures

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The dusky wireworm, *Agriotes obscurus* L. (Coleoptera: Elateridae) is a significant pest of vegetable and field crops in the Fraser Valley of BC (Vernon *et al.* 2001). Adults emerge from the soil from late March through May and die soon after mating (males) or egg laying (Brian 1947), restricting their availability for research and necessitating storage methods that prolong survival. Here we compare beetle survival during and after storage at various temperatures for various durations.

Male *A. obscurus* beetles were collected at the Pacific Agri-Food Research Centre (Agassiz, BC) during their peak emergence (1st wk of May, 2005) and placed in groups of 10 beetles into 150 ml plastic containers with a freshly-cut apple piece (approx. 2 x 2 x 0.5cm) placed on 70 g of moist sandy clay-loam soil. Containers were put in growth chambers (15 per chamber) (Controlled Environments Ltd., Winnipeg, MB) set at 5, 8.5, 12, and 20 °C (± 0.1 °C), and dead beetles removed, and apple pieces replaced, biweekly. Three containers were removed from each chamber after 2, 4, 6, 8, and 10 wks to determine subsequent survival at room temperature (RT) (21 ±1 ° C), and beetles transferred to 10cm Petri dishes (one per container) placed on 5cm high racks inside Styrofoam boxes. Each box (36 x 26 x 9cm deep) contained 2.5ml water to prevent desiccation; a 1cm gap between the box and its lid permitted air exchange. Beetle feeding and observation continued (as above) for up to 12 wks.

Beetle mortality during storage was highest at 20 °C, and similar at 5, 8.5, and 12° C for the first 6 wks but considerably lower at 8.5 °C thereafter. Beetle mortality was rapid within the first 2 wks of storage, and increased with duration for all temperatures except 8.5 °C (Table 1). Regression (stepwise, backward; Proc REG, SAS 9.1; SAS Institute 2002) of the proportion dead per container during storage (m) to temperature (t) and duration (d) yielded the following: $m = 0.003t^2 - 0.022t + 0.050d$ (SE slopes: 0.0005, 0.012, 0.009 respectively; $P < 0.0001$, 0.06, <0.0001 respectively, d.f. = 3,57, adj. $R^2 = 0.88$), indicating that survival increases as storage temperature decreases.

For post-storage survival analysis, each beetle was considered an experimental unit, and storage temperatures were compared with Kaplan-Meier survival analysis (Proc LIFETEST, SAS 9.1), with strata duration (Cox and Oakes 1984). The survival time of 50% of beetles (ST50) was subsequently estimated by modelling survivorship for each storage temperature-duration combination. Survivorship curves were compared with log-rank tests; ST50 values were compared using 95% confidence intervals.

Post-storage beetle mortality was rapid regardless of previous storage temperature. Beetles stored at 12 °C died more quickly at RT than those stored at 8.5 or 5 °C ($\chi^2 = 12.64$, $P = 0.0004$; $\chi^2 = 19.27$, $P < 0.0001$, respectively). Beetles stored at 5 °C survived longest, but not significantly longer than those stored at 8.5 °C ($\chi^2 = 2.91$, $P = 0.09$). Comparison of ST50 values and survival curves indicated that beetles survived longer if stored at 5 °C than at 8.5 °C, if

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Table 1.

Mortality of *Agriotes obscurus* beetles after storage at various temperature for different durations, and subsequent survivorship at room temperature (RT). N = 30 per storage duration-temperature combination. Post-storage survivorship at RT modelled with Kaplan-Meier survival curves; ST50 = time required for 50% survival.

Storage temperature (°C)	Storage duration (wk)	Mean (SE) number of beetles dead (out of 10) at the end of the storage period	Survival of beetles at RT after storage (d)		
			ST50	Upper 95% CI	Lower 95% CI
5	2	2.33 (0.67)	68.0	54.0	89.0
5	4	2.0 (0.58)	20.0	12.0	33.0
5	6	3.0 (1.53)	19.0	5.0	33.0
5	8	4.0 (2.08)	30.5	19.0	47.0
5	10	5.67 (0.33)	12.0	5.0	26.0
8.5	2	2.0 (0.58)	27.0	26.0	47.0
8.5	4	2.0 (1.0)	26.0	14.0	47.0
8.5	6	3.67 (0.33)	19.0	14.0	33.0
8.5	8	1.33 (0.67)	19.0	12.0	21.0
8.5	10	2.67 (1.67)	12.0	7.0	33.0
12	2	1.33 (0.88)	33.0	12.0	47.0
12	4	2.67 (0.33)	12.0	12.0	14.0
12	6	3.00 (0.58)	12.0	5.0	21.0
12	8	4.67 (2.19)	9.5	5.0	19.0
12	10	8.67 (0.88)	n/a[1]	n/a[1]	n/a[1]
20	2	5.0 (1.15)	5.0	5.0	7.0
20	4	9.67 (0.33)	n/a[1]	n/a[1]	n/a[1]
20	6, 8, 10	10.0 (0)	n/a[1]	n/a[1]	n/a[1]

[1] Not enough beetles survived storage to permit analysis.

stored for 2 wks ($\chi^2 = 15.48$, $P<0.0001$) or 8 wks ($\chi^2 = 6.15$, $P=0.013$; Table 1), but not when stored for 4 or 6 wks ($P>0.05$, Table 1). Surprisingly, beetles stored for 2 wks at 5, 8.5, or 12 °C survived longer at RT than those stored at 20 °C (Table 1).

These results indicate that storage at lower temperatures prolongs male click beetle survival, and that storage at 8.5 °C caused highest overall survival. Future research should investigate how cold storage conditions affects beetle physiology.

We thank Drs. D. Gray, C. Stevenson, D. Raworth and R. Bennett, and sundry reviewers for valuable advice.

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